

PCTWORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁷ : G01N 15/14	A1	(11) International Publication Number: WO 00/54026 (43) International Publication Date: 14 September 2000 (14.09.00)
(21) International Application Number: PCT/DK00/00092 (22) International Filing Date: 6 March 2000 (06.03.00) (30) Priority Data: PA 1999 00317 5 March 1999 (05.03.99) DK (71) Applicants (for all designated States except US): HATTING-KS [DK/DK]; Oensvej 48, Hatting, DK-8700 Horsens (DK). DE DANSKE KVÆGAFLSFØRENINGER [DK/DK]; Udkærsvvej 15, Skejby, DK-8200 Århus N (DK). (72) Inventors; and (75) Inventors/Applicants (for US only): CHRISTENSEN, Preben [DK/DK]; Dalgas Boulevard 89, 1.th., DK-2000 Frederiksberg (DK). STENVANG, Jens, Peter [DK/DK]; Bransbjergvej 119, DK-2600 Glostrup (DK). (74) Agent: PLOUGMANN, VINGTOFT & PARTNERS A/S; Sankt Annæ Plads 11, P.O. Box 3007, DK-1021 Copenhagen K (DK).		(81) Designated States: AE, AL, AM, AT, AT (Utility model), AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, CZ (Utility model), DE, DE (Utility model), DK, DK (Utility model), DM, EE, EE (Utility model), ES, FI, FI (Utility model), GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SK (Utility model), SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG). Published <i>With international search report.</i>
(54) Title: DETERMINATION OF SPERM CONCENTRATION AND VIABILITY FOR ARTIFICIAL INSEMINATION		
(57) Abstract <p>A method for the simultaneous determination of the total concentration of sperm cells and the proportion of live sperm cells in a semen sample is provided. By selective staining of live, dead and/or dying sperm cells, for example using one or more fluorochromes, the detection means can distinguish between live, dead and/or dying sperm cells, for example by detection of the fluorescent response from the sperm cells. The selective staining may be performed at room temperature and in concentrations below standard concentrations. The determination may be performed by selective counting of cells of each fluorescent quality for example by using a flow cytometer having internal concentration standard means. Furthermore, the likelihood of fertilizing a female animal with an insemination dose, taken from a semen sample having a known total concentration of sperm cells and a known proportion of live sperm cells, is predicted on the basis of the thus determined total concentration of sperm cells and the proportion of live sperm cells.</p>		

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Swaziland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BA	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Barbados	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav Republic of Macedonia	TM	Turkmenistan
BF	Burkina Faso	GR	Greece	ML	Mali	TR	Turkey
BG	Bulgaria	HU	Hungary	MN	Mongolia	TT	Trinidad and Tobago
BJ	Benin	IE	Ireland	MR	Mauritania	UA	Ukraine
BR	Brazil	IL	Israel	MW	Malawi	UG	Uganda
BY	Belarus	IS	Iceland	MX	Mexico	US	United States of America
CA	Canada	IT	Italy	NE	Niger	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NL	Netherlands	VN	Viet Nam
CG	Congo	KE	Kenya	NO	Norway	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NZ	New Zealand	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's Republic of Korea	PL	Poland		
CM	Cameroon	KR	Republic of Korea	PT	Portugal		
CN	China	KZ	Kazakhstan	RO	Romania		
CU	Cuba	LC	Saint Lucia	RU	Russian Federation		
CZ	Czech Republic	LI	Liechtenstein	SD	Sudan		
DE	Germany	LK	Sri Lanka	SE	Sweden		
DK	Denmark	LR	Liberia	SG	Singapore		
EE	Estonia						

1/7

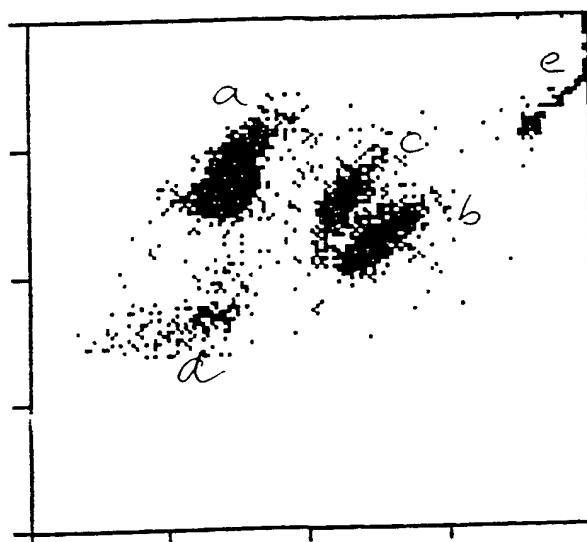


Fig. 1

2/7

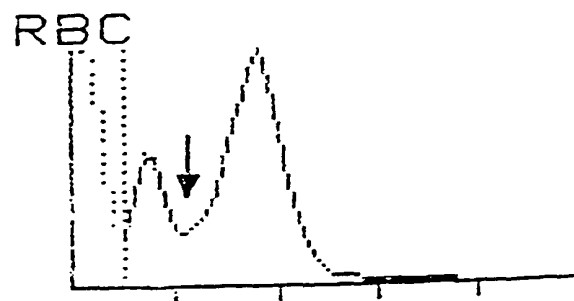


Fig. 2A

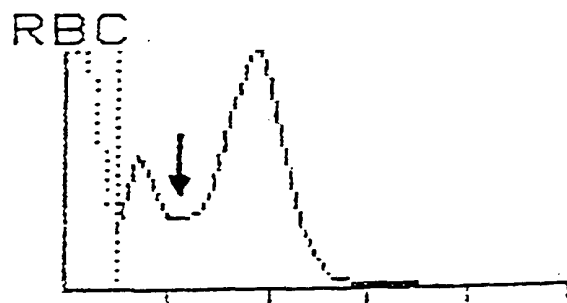


Fig. 2B

3/7

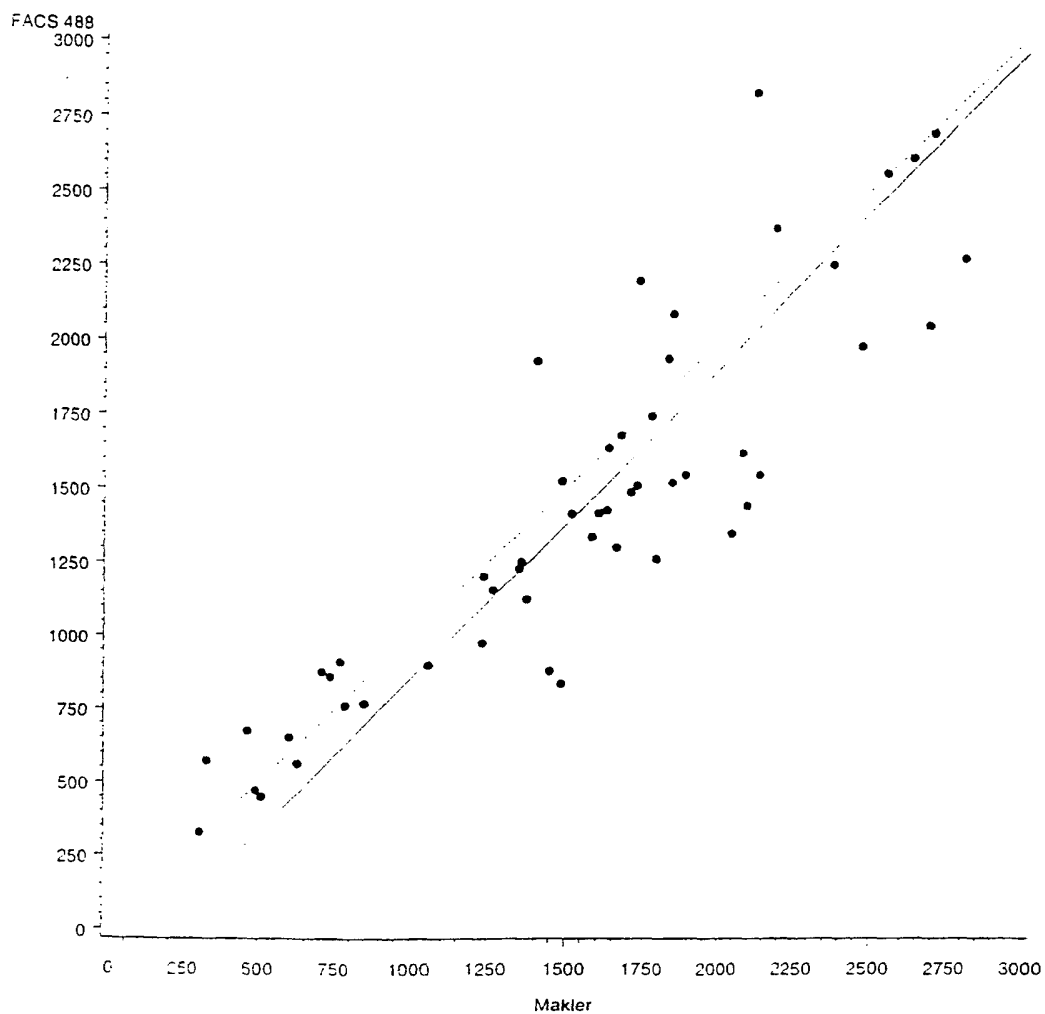


Fig. 3

4/7

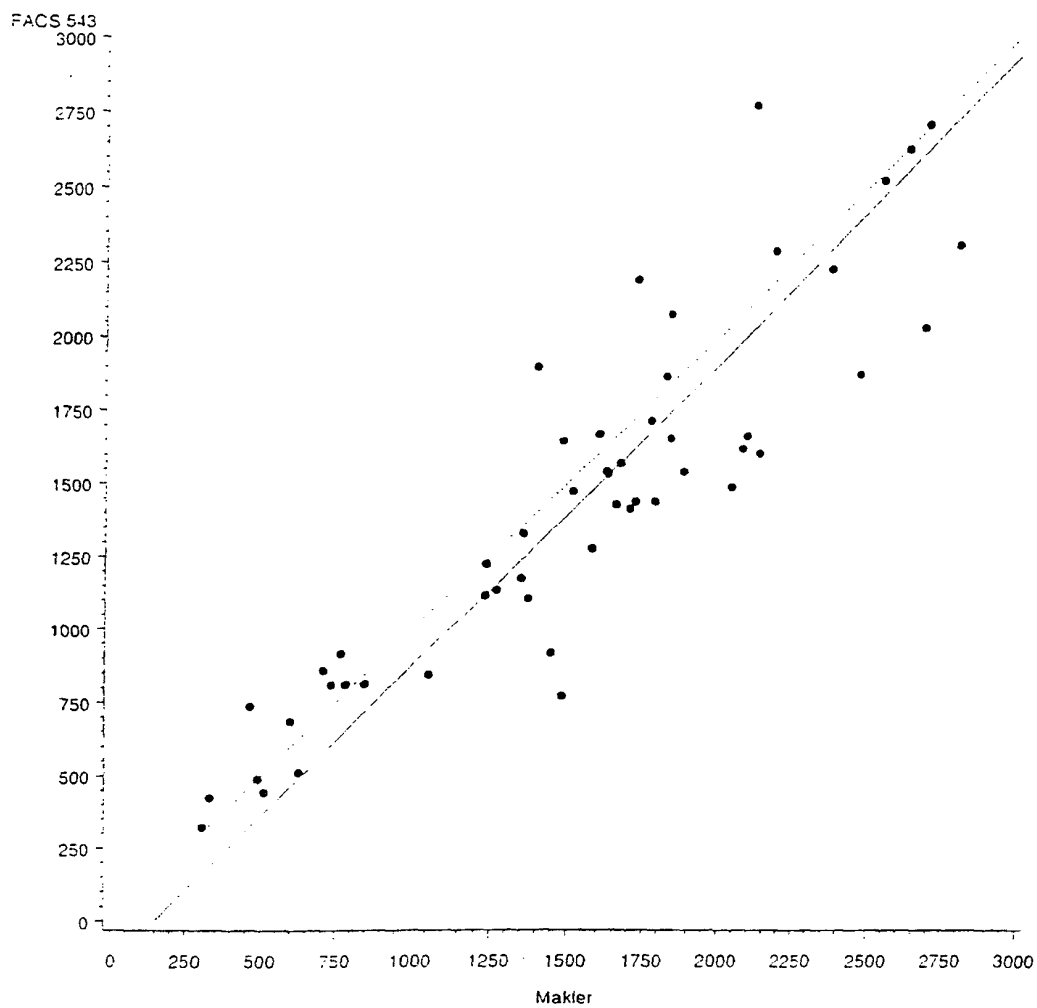


Fig. 4

5/7

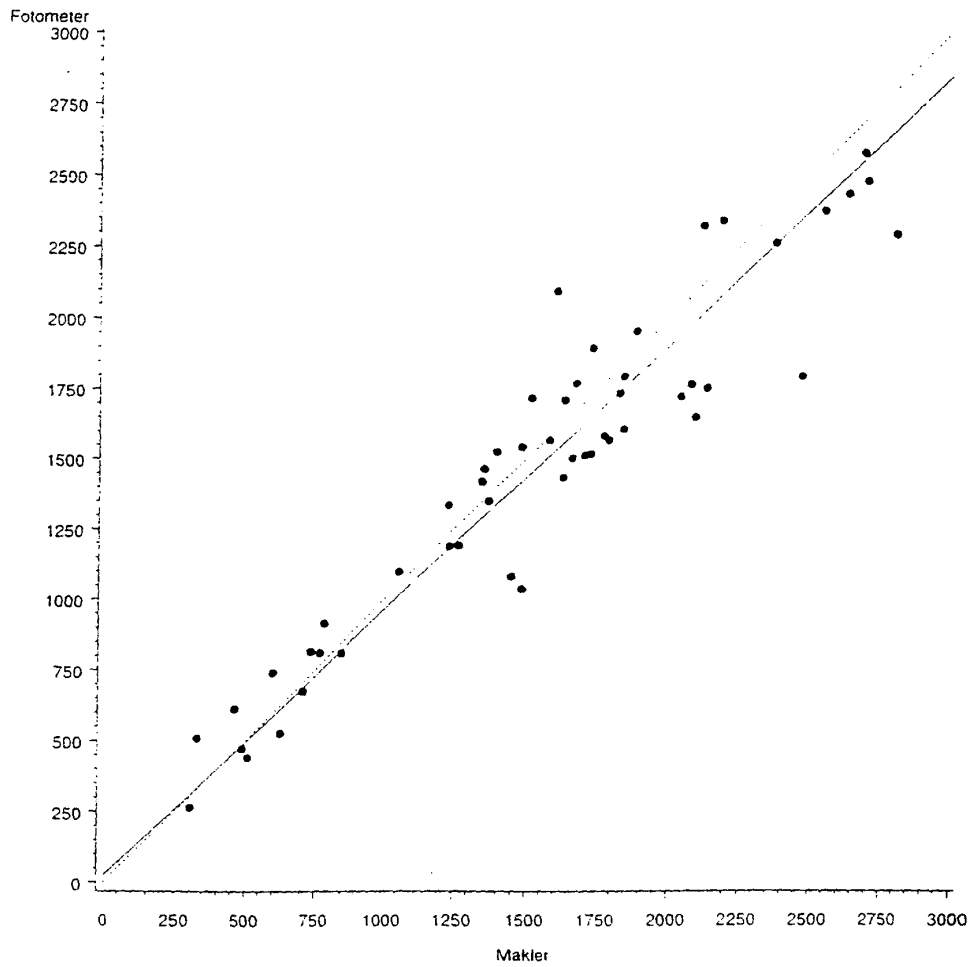


Fig. 5

6/7

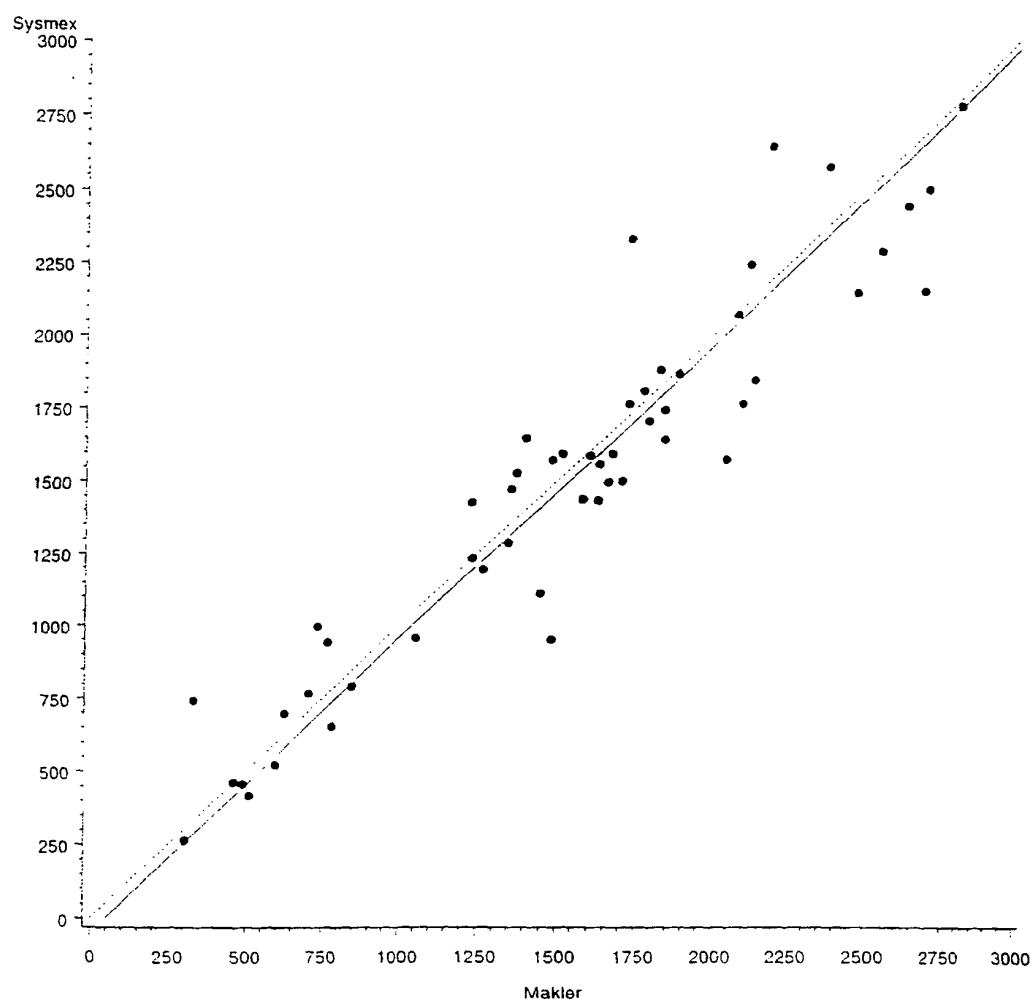


Fig. 6

7/7

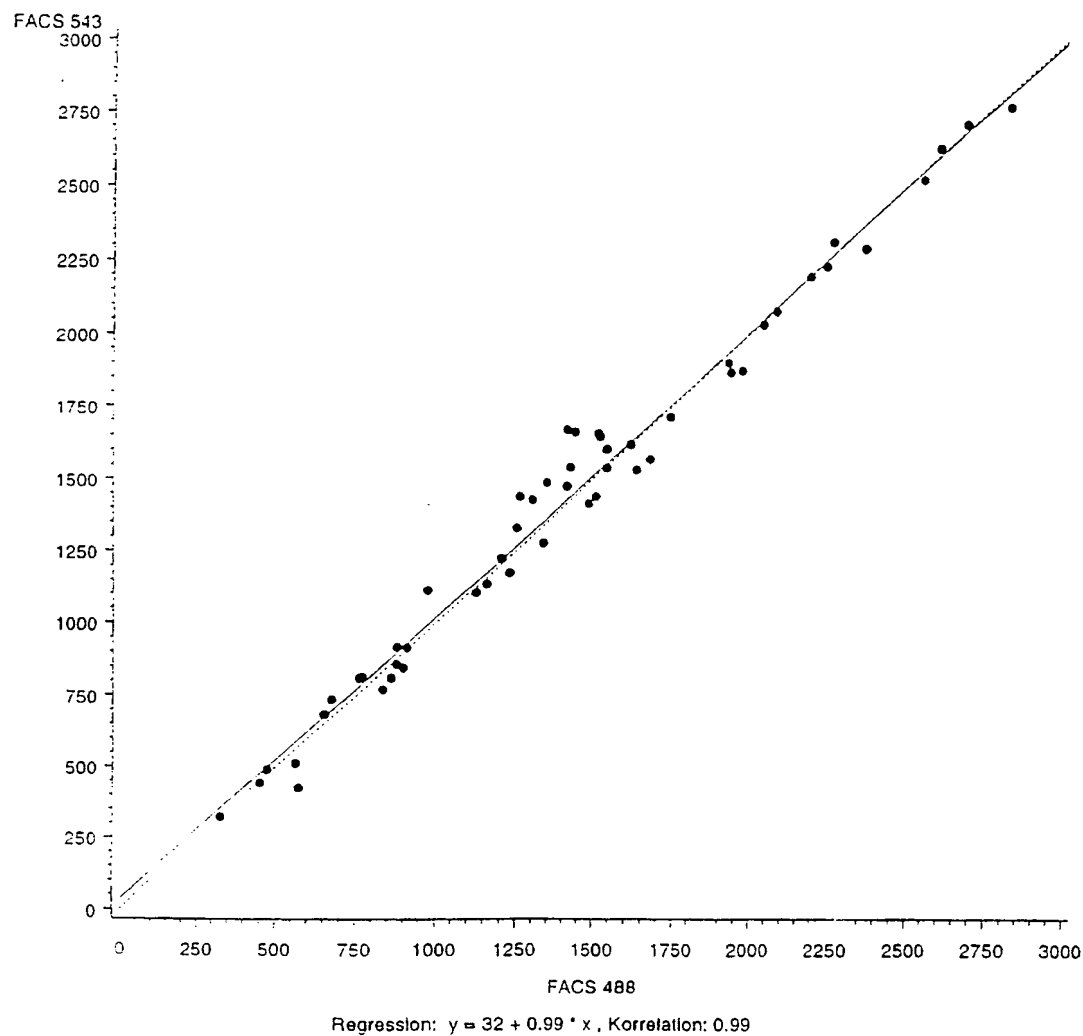


Fig. 7

INTERNATIONAL SEARCH REPORT

International Application No

PCT/DK 00/00092

A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 G01N15/14

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 G01N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	<p>GB 2 214 518 A (INNOFINANCE ALTALANOS INNOVACIOS PENZINTEZET) 6 September 1989 (1989-09-06) figure 1 page 7, line 5 -page 10, line 21 page 5, line 9 -page 6, line 4 page 3, line 10 - line 25 page 2, line 12 - line 15 page 1, line 4 - line 14 --- -/--</p>	1,7-10, 33

☒ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search

14 June 2000

Date of mailing of the international search report

23/06/2000

Name and mailing address of the ISA

European Patent Office, P.B. 5818 Patentlaan 2
NL - 2280 HV Rijswijk
Tel. (+31-70) 340-2040, Tx. 31 651 epo nl,
Fax: (+31-70) 340-3016

Authorized officer

Thomas, R.M.

INTERNATIONAL SEARCH REPORT

International Application No

PCT/DK 00/00092

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	<p>US 4 751 188 A (VALET) 14 June 1988 (1988-06-14)</p> <p>abstract column 2, line 4 - line 11 column 2, line 28 - line 35 column 2, line 44 - line 57 column 3, line 7 - line 29 column 5, line 27 - line 57 column 6, line 11 - line 21 column 7, line 54 - column 8, line 4 figure 1</p> <p style="text-align: center;">---</p>	<p>1-12, 17-24, 26-28</p>
Y	<p>US 4 559 309 A (EVENSON) 17 December 1985 (1985-12-17) cited in the application abstract column 3, line 43 - column 4, line 21 column 4, line 58 - line 65 column 5, line 6 - line 12 column 5, line 34 - line 47 column 5, line 65 - line 66</p> <p style="text-align: center;">---</p>	<p>1-12, 17-24, 26-28</p>
A	<p>US 5 691 133 A (CRITSER) 25 November 1997 (1997-11-25)</p> <p>column 8, line 4 - line 9 column 8, line 15 - line 36</p> <p style="text-align: center;">---</p>	<p>1,5-9, 11,12, 24,29,30</p>
A	<p>GB 2 145 112 A (MILK MARKETING BOARD) 20 March 1985 (1985-03-20) abstract page 2, line 41 - line 50 page 4, line 8 - line 21</p> <p style="text-align: center;">---</p>	<p>1,10, 17-20,24</p>
A	<p>PATENT ABSTRACTS OF JAPAN vol. 1997, no. 4, 30 April 1997 (1997-04-30) & JP 08 332098 A (ORIENTAL YEAST), 17 December 1996 (1996-12-17) abstract</p> <p style="text-align: center;">---</p>	<p>1,10,11</p>
A	<p>US 5 812 312 A (LORINCZ) 22 September 1998 (1998-09-22) column 2, line 41 - line 47 * column 3, penultimate line - column 4, line 18 * column 4, line 32 - line 44 column 5, line 51 - line 53 figure 4</p> <p style="text-align: center;">---</p>	<p>1,7-10, 17-20</p>
A	<p>WO 93 16385 A (COULTER CORPORATION) 19 August 1993 (1993-08-19) claims 1,4</p> <p style="text-align: center;">---</p>	<p>1,21-23</p>
	-/--	

INTERNATIONAL SEARCH REPORT

International Application No.

PCT/DK 00/00092

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	EP 0 586 183 A (BECTON DICKINSON) 9 March 1994 (1994-03-09) claim 1 ---	1,21-23
A	US 5 229 265 A (TOMETSKO) 20 July 1993 (1993-07-20) column 3, line 52 - line 56 column 7, line 36 - line 45 column 20, line 59 - line 65 column 22, line 22 - line 41 ---	1,21-23
A	US 5 866 354 A (FROMAN) 2 February 1999 (1999-02-02) column 1, paragraph 1 - paragraph 2 figure 11; example 7 ---	33-37, 40,41
A	WO 95 29983 A (PENN STATE) 9 November 1995 (1995-11-09) page 1, paragraph 1 - paragraph 3 examples 3,10,11; table 1 -----	33,37,38

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/DK 00/00092

Patent document cited in search report		Publication date	Patent family member(s)	Publication date
GB 2214518	A	06-09-1989	AU 596270 B	26-04-1990
			AU 1118588 A	03-08-1989
			CH 675485 A	28-09-1990
			DE 3806556 A	07-09-1989
			ES 2006560 A	01-05-1989
			FR 2626979 A	11-08-1989
			JP 1210863 A	24-08-1989
			NL 8800239 A	01-09-1989
			SE 460121 B	11-09-1989
			SE 8800325 A	03-08-1989
			US 4880732 A	14-11-1989
US 4751188	A	14-06-1988	DE 3238353 A	19-04-1984
			CA 1219791 A	31-03-1987
			EP 0106339 A	25-04-1984
			JP 60022661 A	05-02-1985
US 4559309	A	17-12-1985	NONE	
US 5691133	A	25-11-1997	US 5595866 A	21-01-1997
			AU 2650795 A	21-12-1995
			US 6054287 A	25-04-2000
			WO 9532624 A	07-12-1995
			US 5700632 A	23-12-1997
			US 5753427 A	19-05-1998
			US 5776769 A	07-07-1998
GB 2145112	A	20-03-1985	NONE	
JP 08332098	A	17-12-1996	NONE	
US 5812312	A	22-09-1998	AU 9376298 A	22-03-1999
			WO 9912057 A	11-03-1999
WO 9316385	A	19-08-1993	US 5451525 A	19-09-1995
			AT 191561 T	15-04-2000
			AU 3617093 A	03-09-1993
			BR 9305886 A	19-08-1997
			CA 2129535 A	15-08-1993
			DE 69328296 D	11-05-2000
			EP 0627080 A	07-12-1994
			FI 943732 A	12-10-1994
			JP 7503795 T	20-04-1995
			NO 942989 A	11-10-1994
EP 0586183	A	09-03-1994	CA 2104901 A	05-03-1994
			DE 69326731 D	18-11-1999
			DE 69326731 T	18-05-2000
US 5229265	A	20-07-1993	NONE	
US 5866354	A	02-02-1999	NONE	
WO 9529983	A	09-11-1995	CA 2188932 A	09-11-1995
			EP 0759065 A	26-02-1997
			JP 9512436 T	16-12-1997
			US 5763206 A	09-06-1998